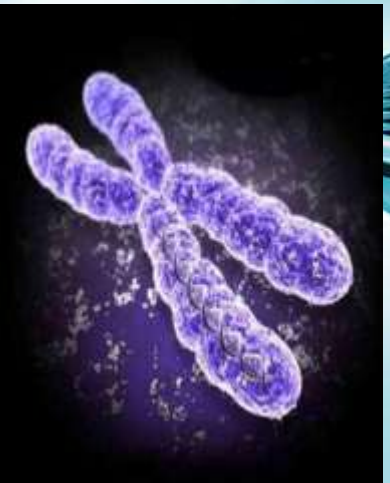
EPIGENETICS AND NEPHROLOGY



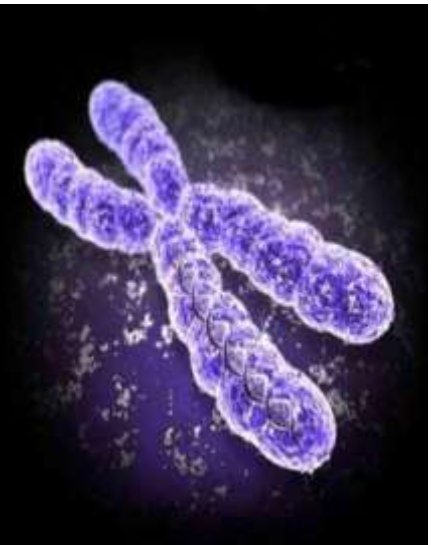
Amr Mohab

Lecturer of Internal Medicine & Nephrology Ain Shams University

2015

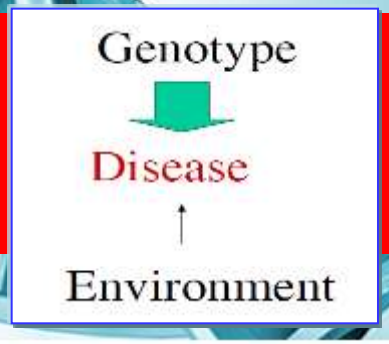
AGENDA

- Basis of genetics
- What is epigenetics?
- Basic epigenetic concepts
- Epigenetic role & applications in Nephrology



Darwin, Mendel, and Lamarck looking sharp portrait style.

Genetics



- Genes control the inheritance of traits **(Mendel, 1865)**
- The human genome project completed in **2003**
- Mutation of a single gene can play a major role in diseases
- Genetics of common, complex traits

will provide risk factors = identify people at higher risk

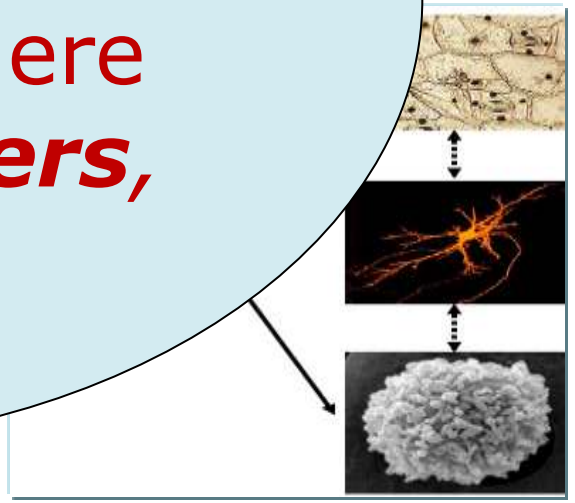
How could you explain?

Identical twins have different natural hair colors?

An Individual with Two Diff

Different adult stem cells know their fate
sts can only form
ls
only form skin
s only
s

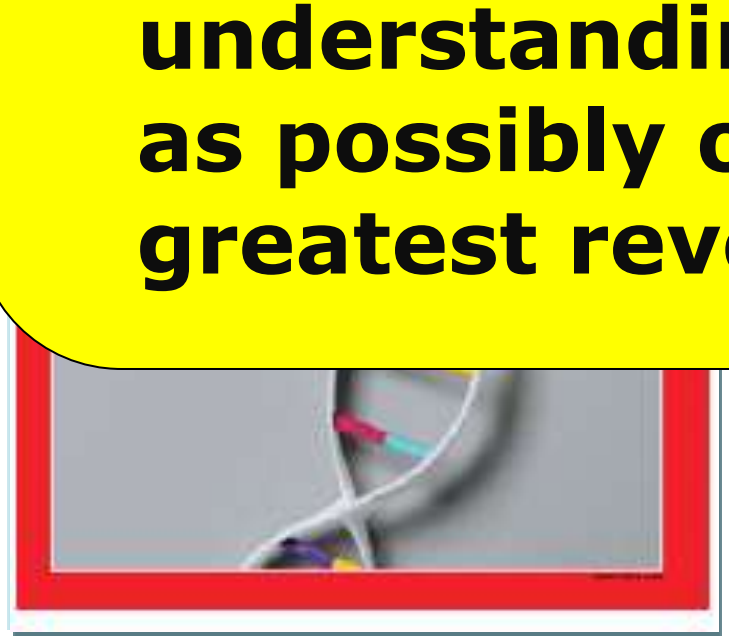
Scientists now know that genes are not the only authors of inheritance. There are ***ghostwriters***, too

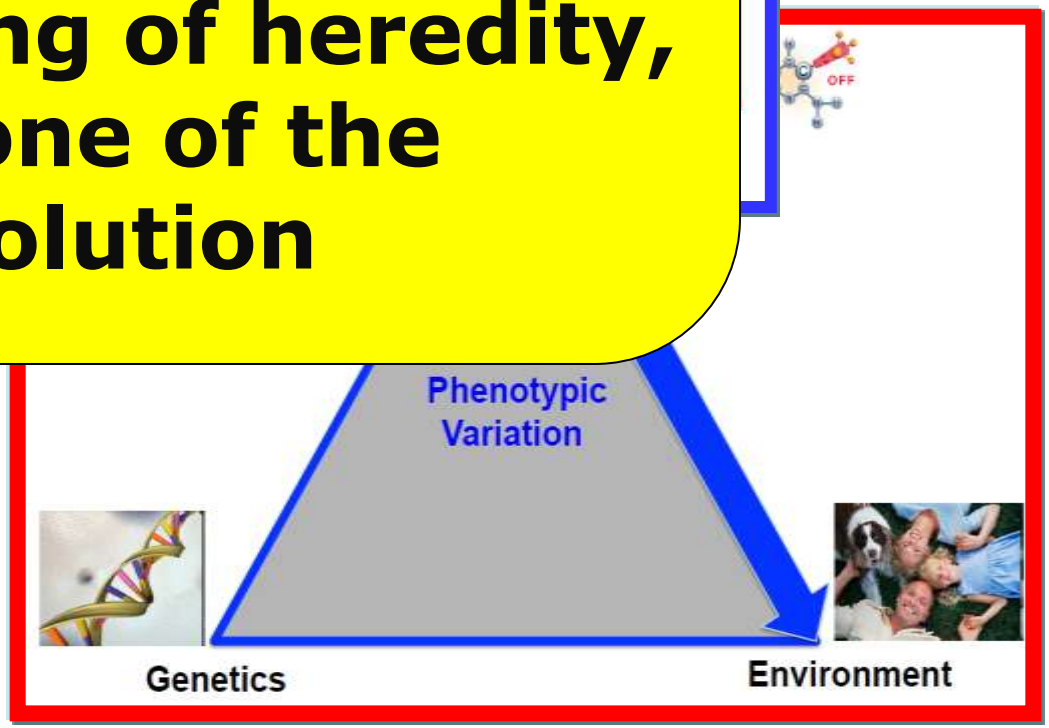


It is Epigenetics not

- ***The rise of epigenetics….*** **in the late 20th century was a turning point in our understanding of heredity, as possibly one of the greatest revolution**

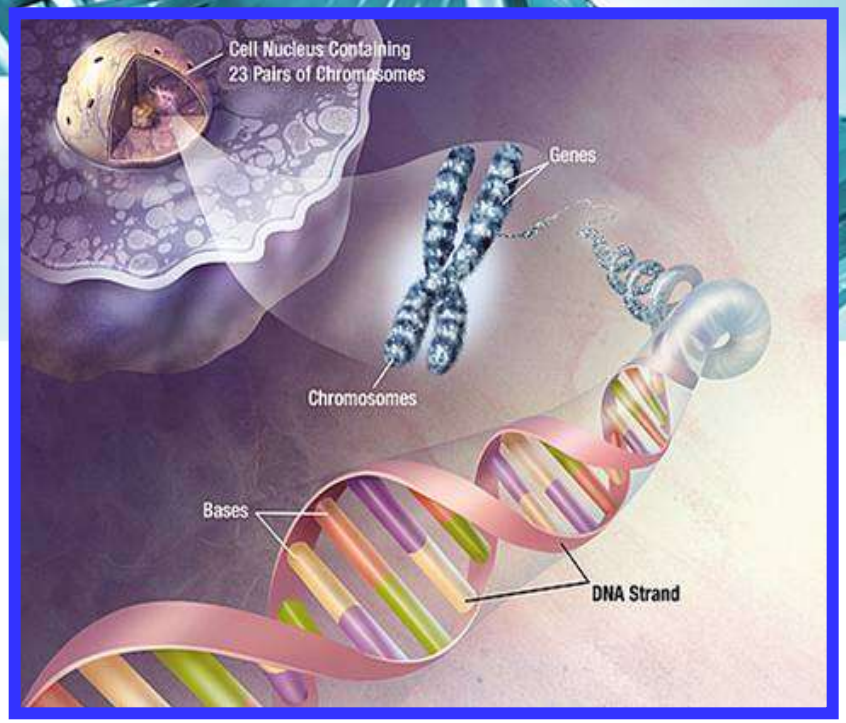
Waddington

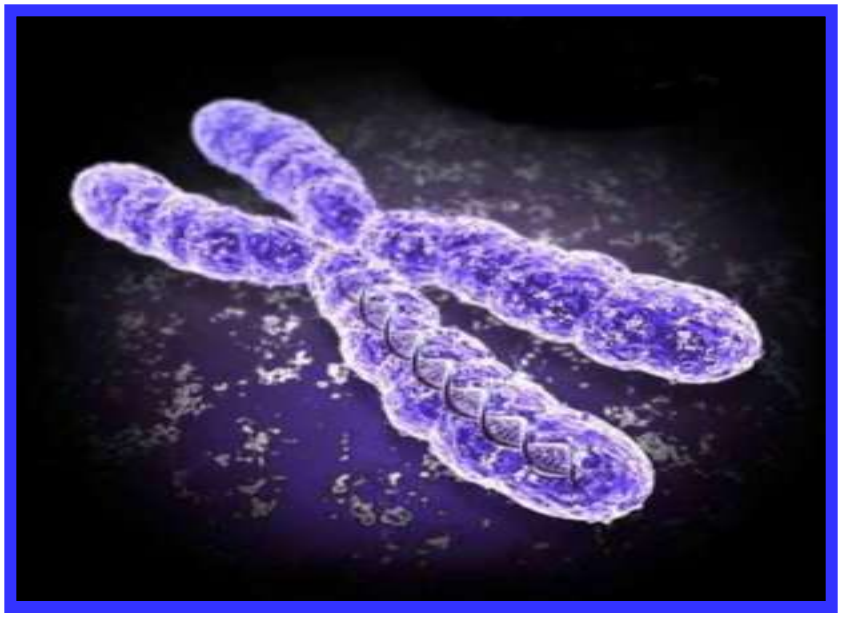




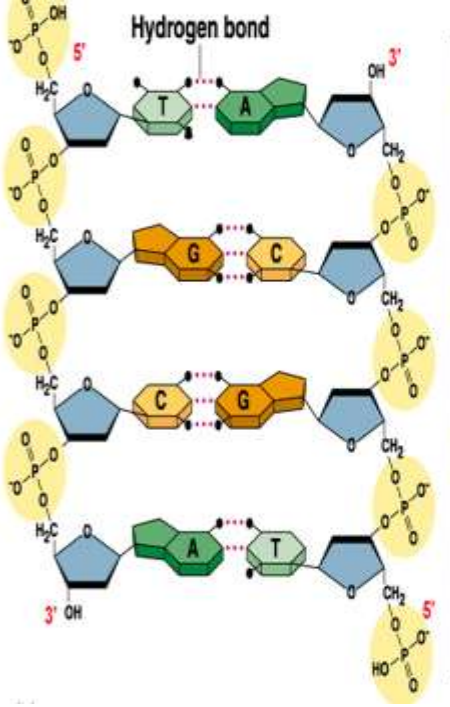
The nucleus

- Chromosomes
- Genes
- DNA





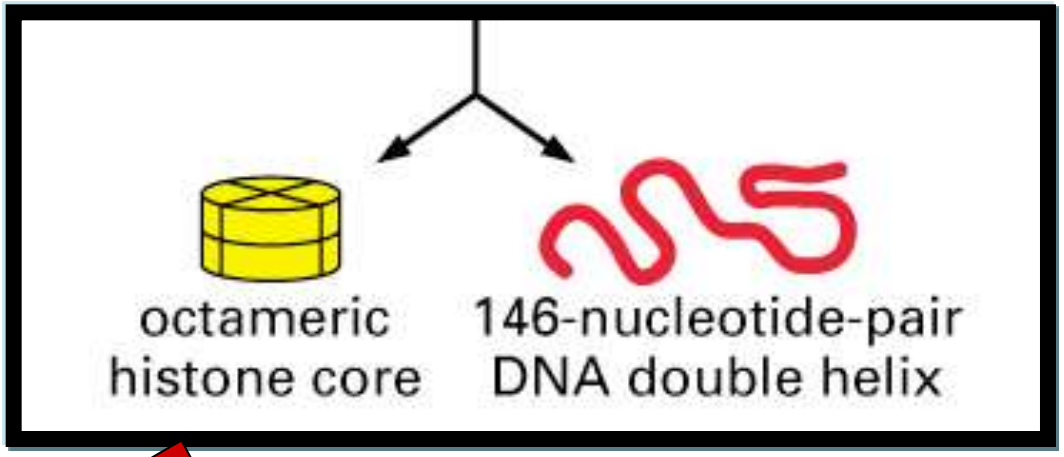
What is DNA?

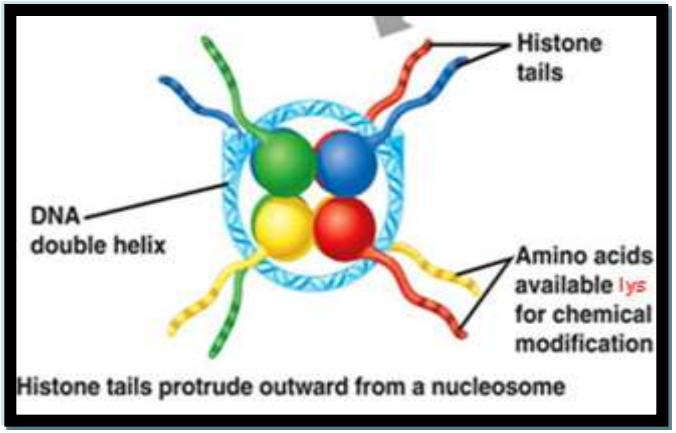


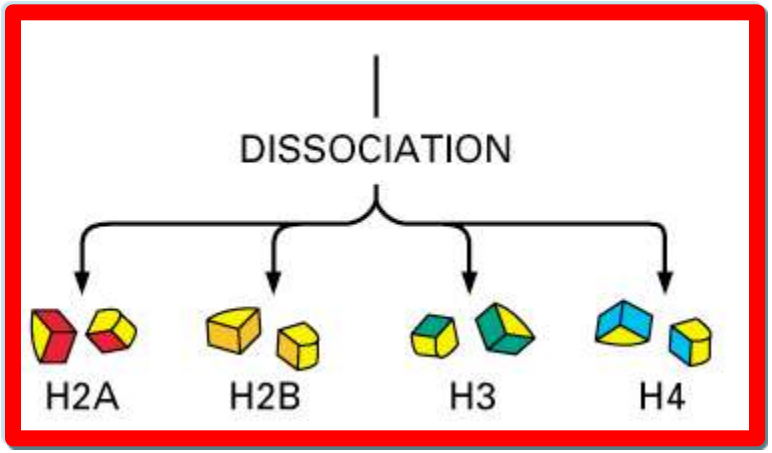
- DNA is a **Double Helix of polyneucleotides**
- Each nucleotide consists of: sugar,PO4 group, nitrogen base (Adenine, Guanine, Thymine, Cytosine)
- **2 m of linear DNA** have to be **packed** into a nucleus of roughly 10 µm diameter
- DNA coils around **8 histone protein cores** forming nucleosomes; the "beads on a string" structure (chromatin).
- Nucleosomes are folded several times to eventually form a **chromosome**

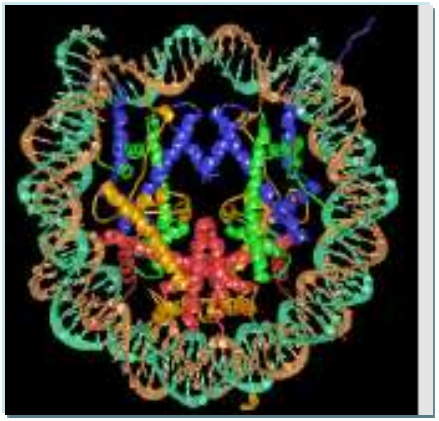
DNA is packaged in nucleosomes

The **nucleosome** → **chromatin fiber**

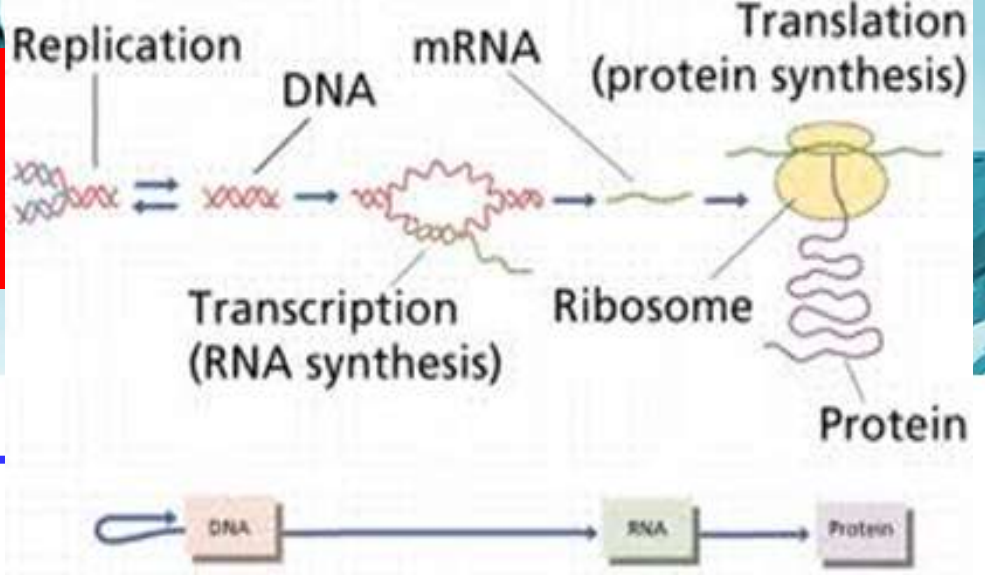








GENETIC CODE

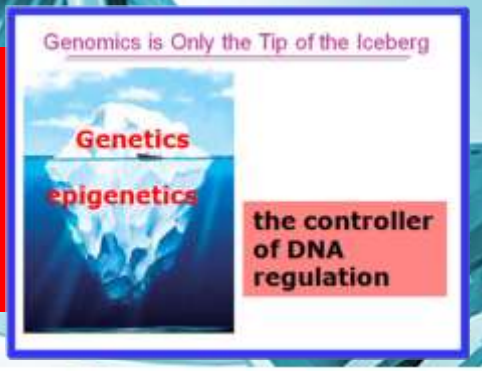


- A gene is a portion of DNA
- The sequence of nucleotides in a gene is read and translated by a cell to produce a chain of amino acids then into proteins this is known as the genetic code
- Conversion of genetic information to a protein relies on the transcription of neculotide sequences of DNA to mRNA

Genes may be activated or inactivated (silenced), determined by a cell's or environment : by interference with transcription by mRNA, or post translation modification

A Silenced Gene is not Transcribed, Translated, or Expressed

Body cell information



- **There are at least 2 forms of information in the genome of the cell:**

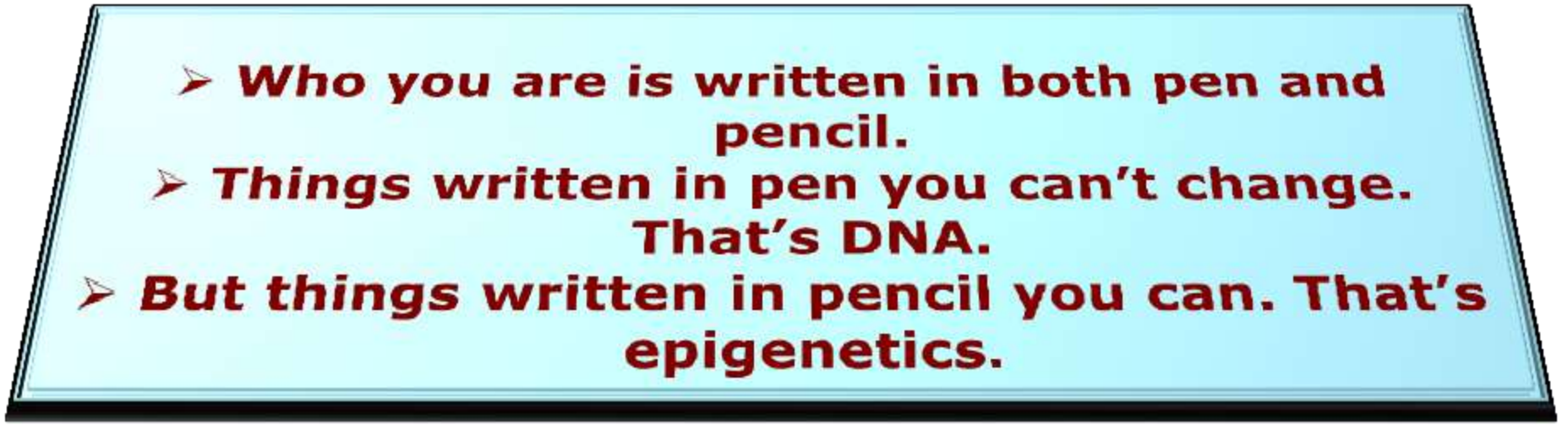
A- **Genetic information: provides the building block for the manufacture of all Proteins needed for the cell functional activity.** Simple genetics based on changes to the DNA sequence (the genotype) *1st Genetic Code*

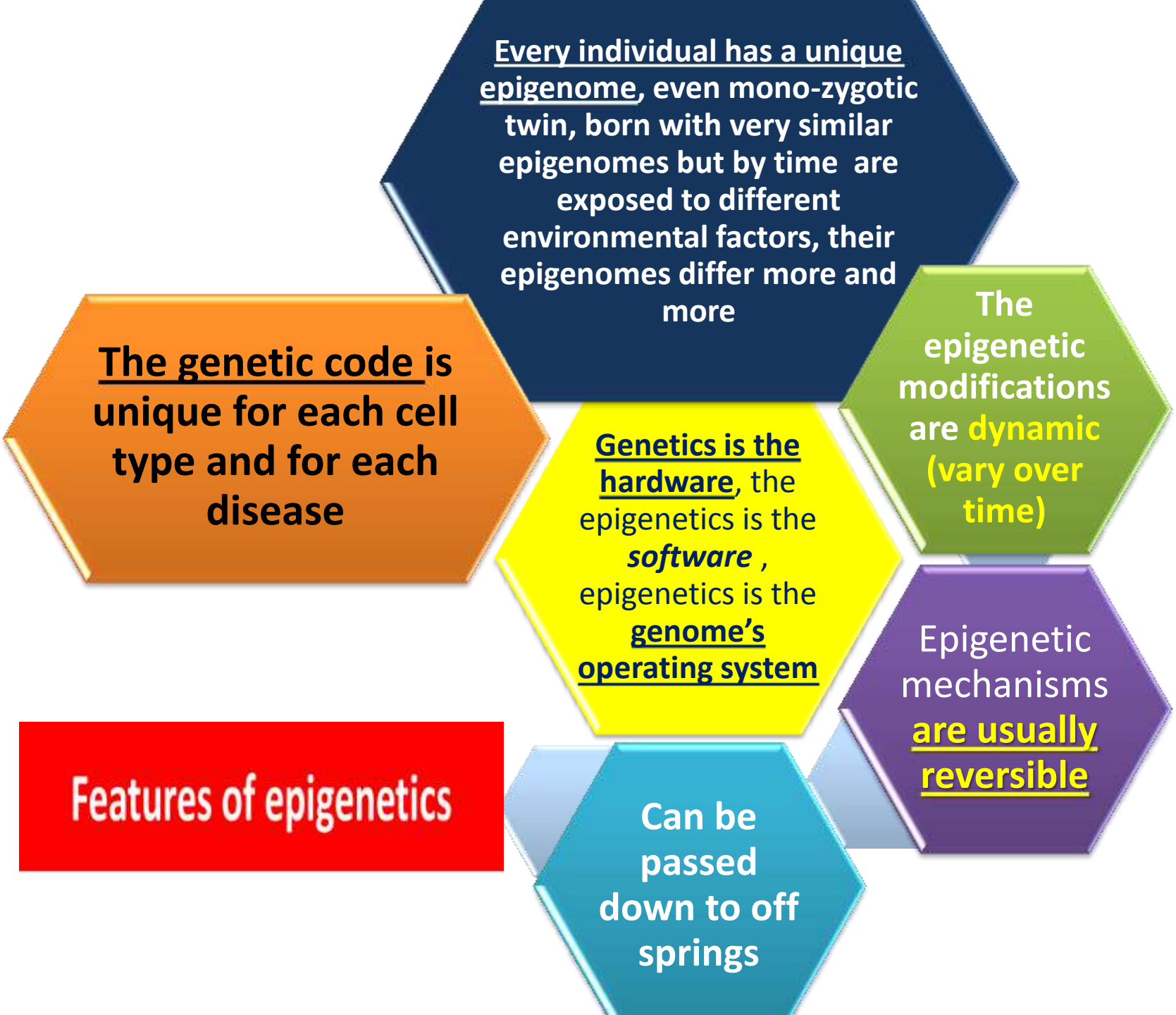
B- **Epigenetic information: provides additional instruction on how, when and where these information should be used,& *decide what happens to genes over course of our life . 2nd Genetic Code***

Marx V. Nature 2012.

What does “epigenetics” mean?
= above or on top of genetics

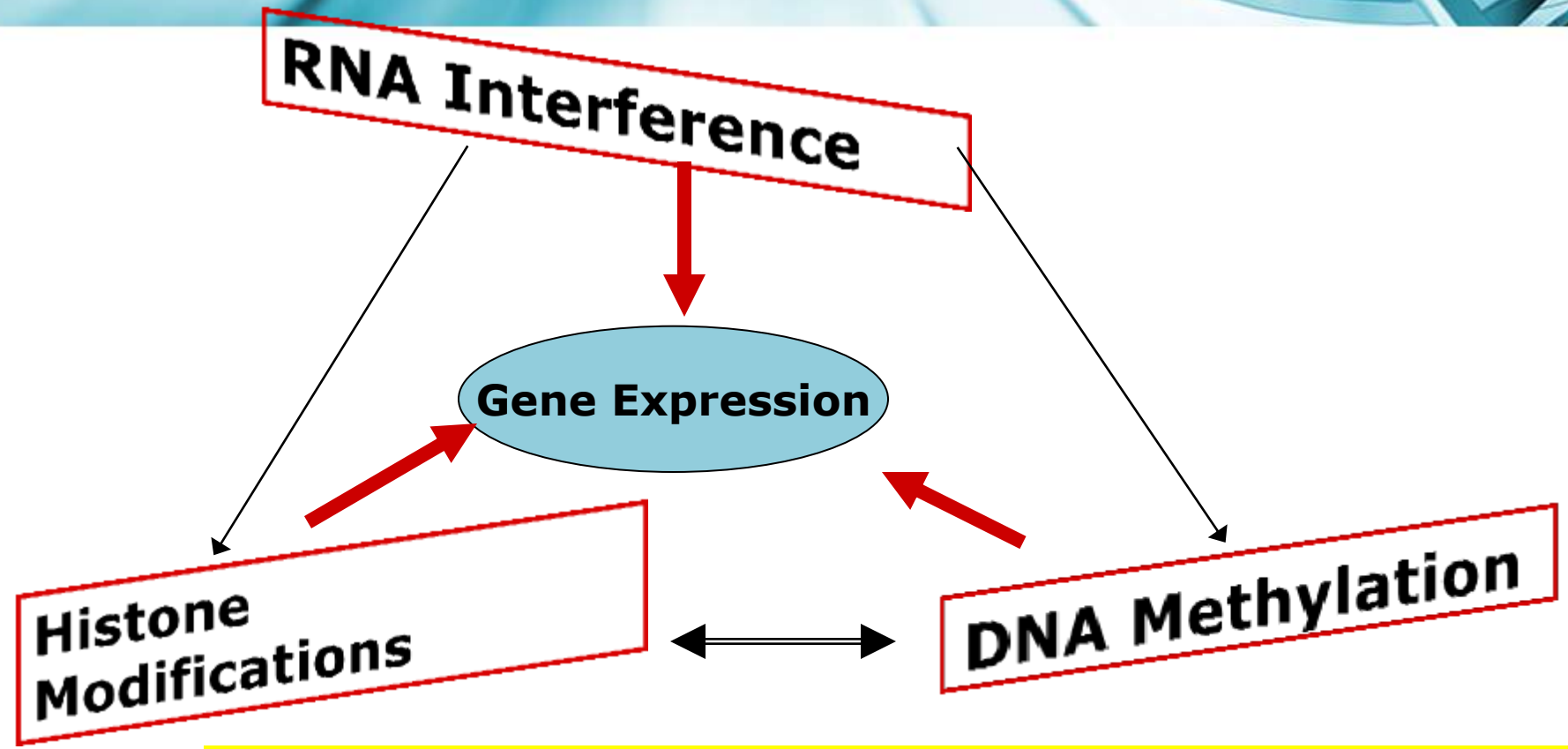
- While epigenetics often refers to the study of single genes or sets of genes, **epigenomics** refers to more global analyses of epigenetic changes across the entire **genome**.




Every individual has a unique epigenome, even mono-zygotic twin, born with very similar epigenomes but by time are exposed to different environmental factors, their epigenomes differ more and more
The genetic code is unique for each cell type and for each disease
The epigenetic modifications are dynamic (vary over time)
Genetics is the hardware, the epigenetics is the software , epigenetics is the genome's operating system
Epigenetic mechanisms are usually reversible
Features of epigenetics
Can be passed down to off springs

How are epigenetic information accomplished ?

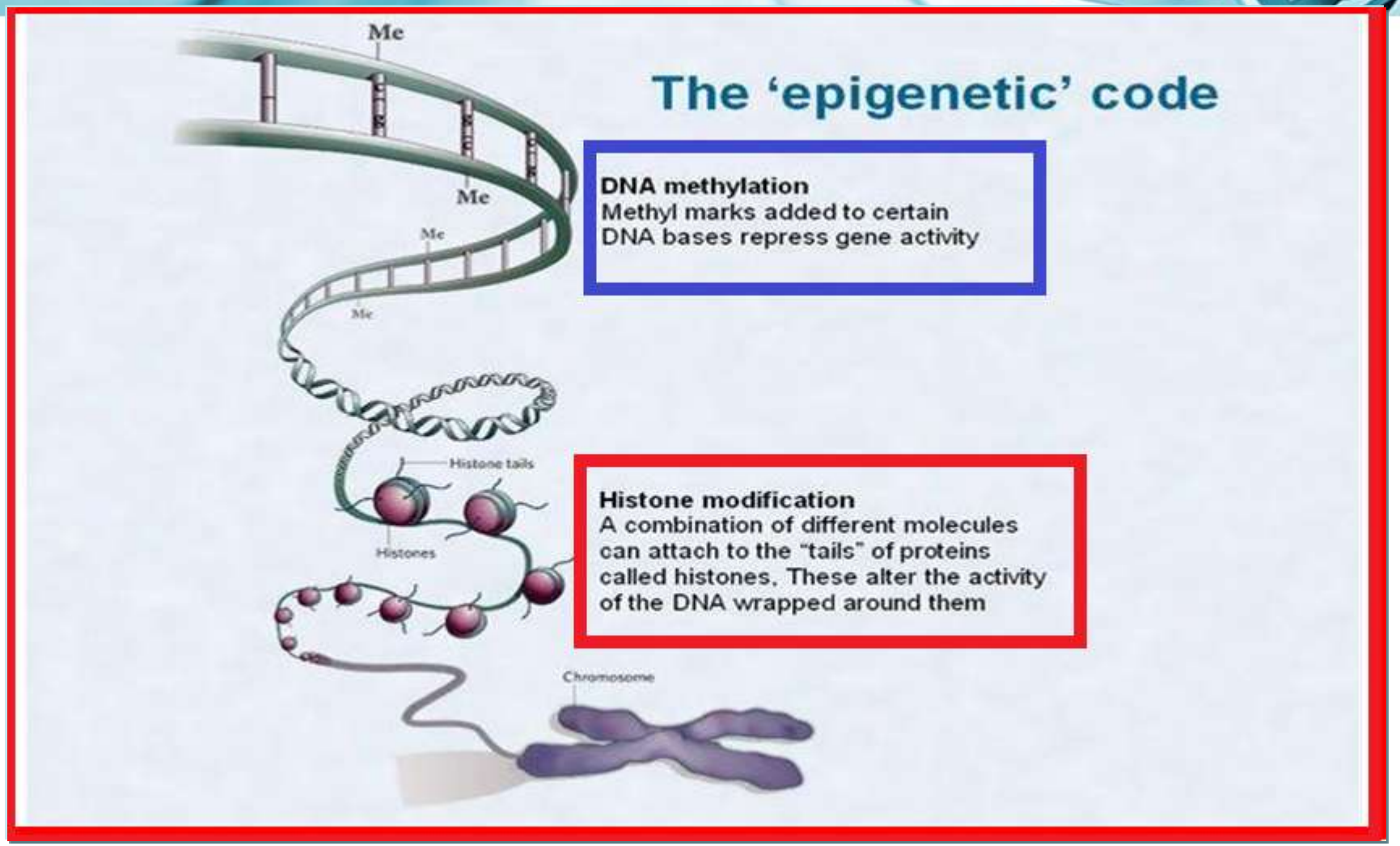
Epigenetics Mechanisms



These heritable modifications are collectively termed

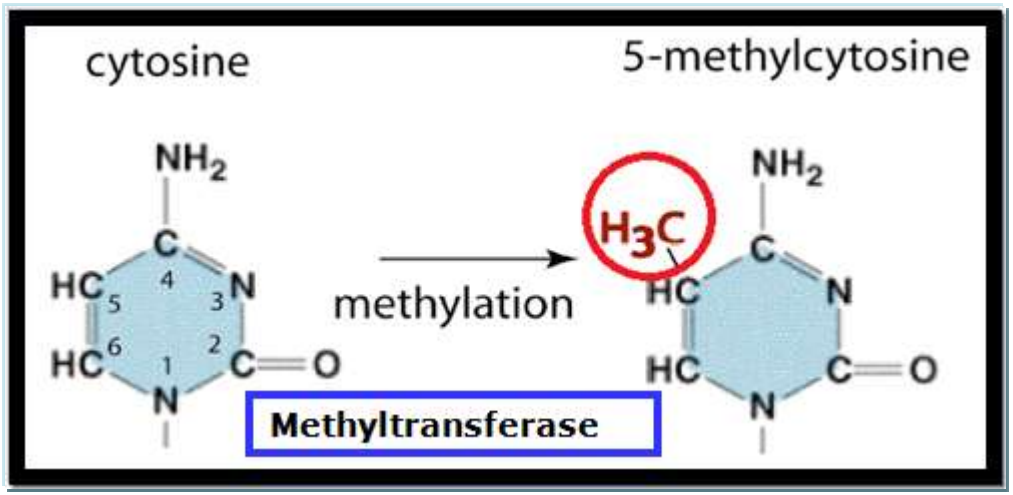
"epigenetic codes"

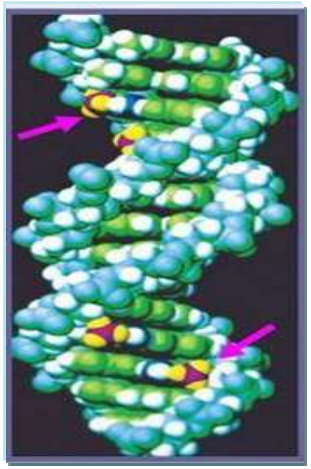
The two main mechanisms of the epigentic code



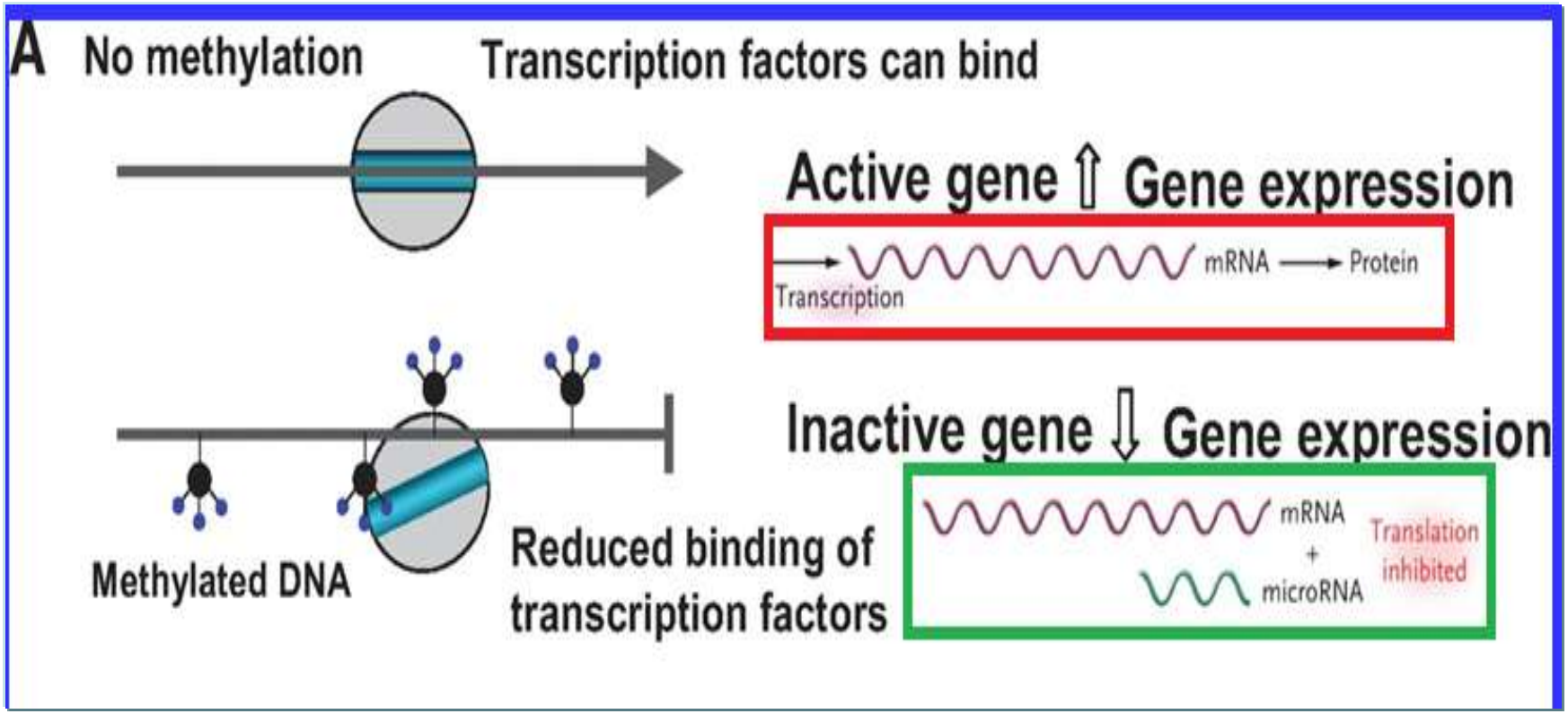
1-DNA Methylation

- Methylation of cytosine in DNA occurs at CpG dinucleotides is a stable **epigentic mark**
- DNA methylation is a modification of **DNA that does not change the DNA sequence**, **but has an influence on gene activity** at the level of transcription or translation





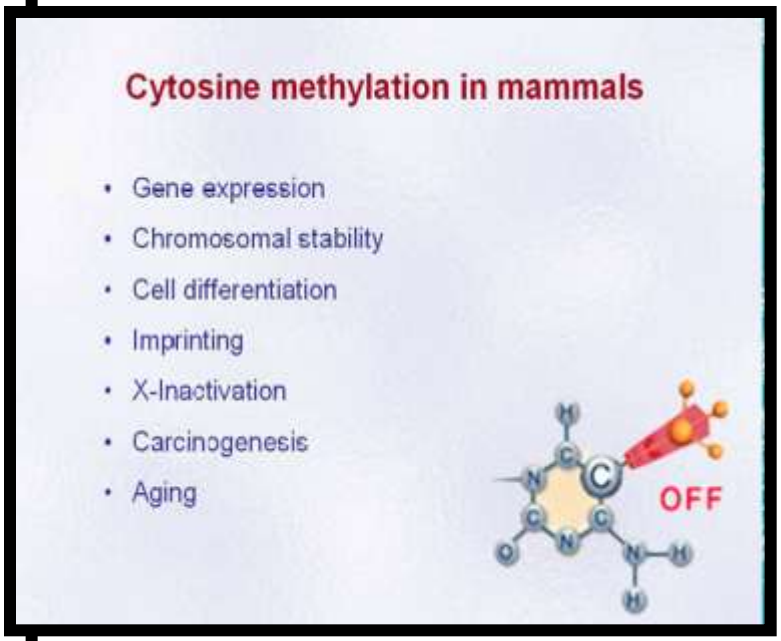
EFFECTS OF DNA METHYLATION ON GENE EXPRESSION



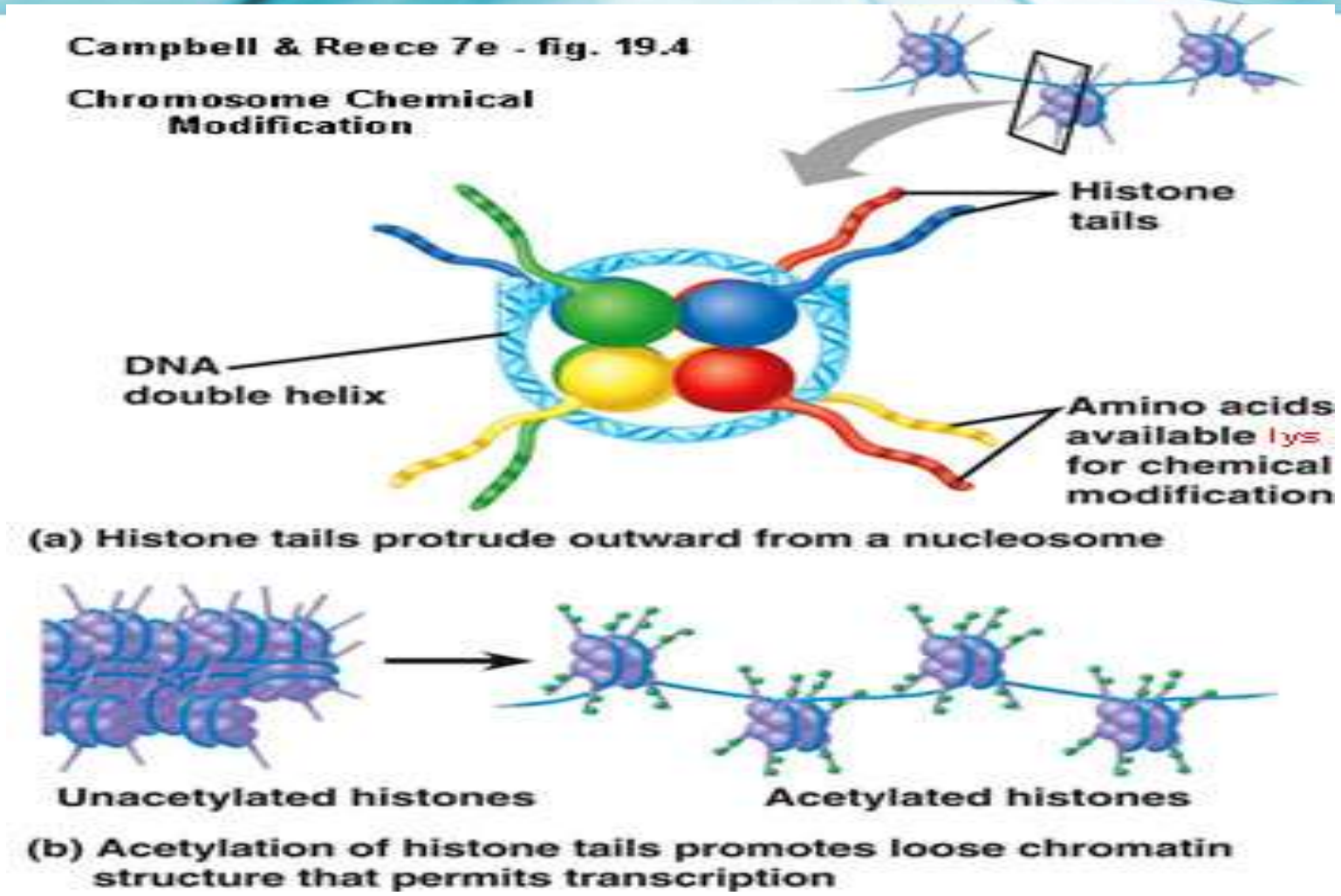
Ling C , and Groop L Diabetes 2009;58:2718-2725

What is the effect of DNA methylation

- Methylation affects **gene activity**, keeps the histones tight together so **the DNA cannot be transcribed/expressed.**
- The presence of 5-methylcytosine leads to **the silencing of genes** in that local area of the chromosome =**gene repression**
- **This change is *reversible*** , making it a ***therapeutic target***

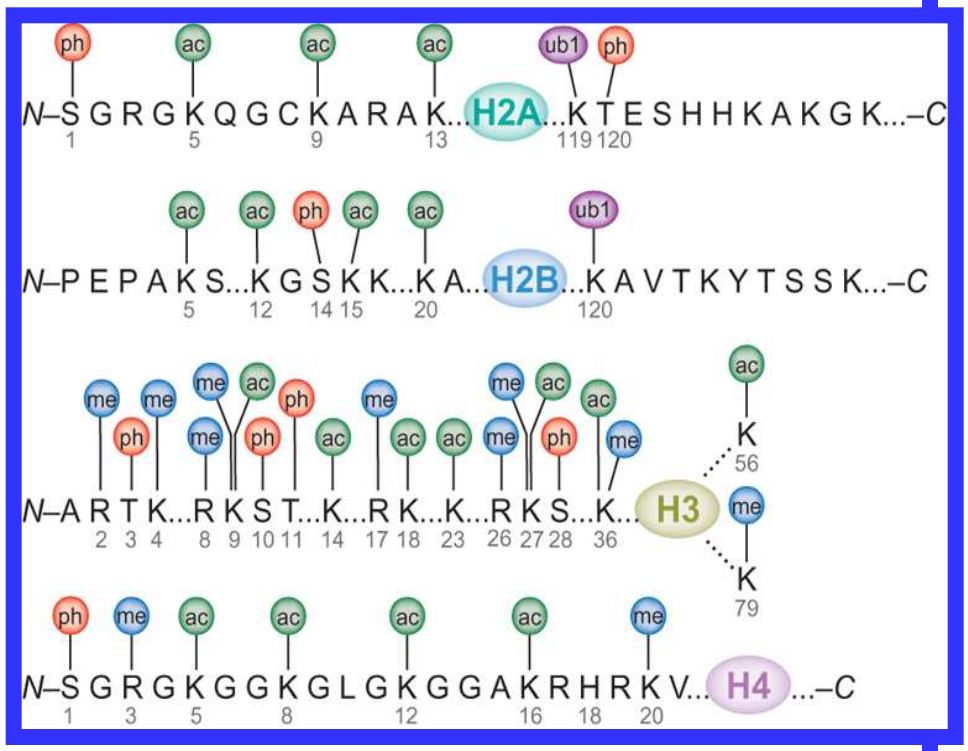


2-Histone Modifications

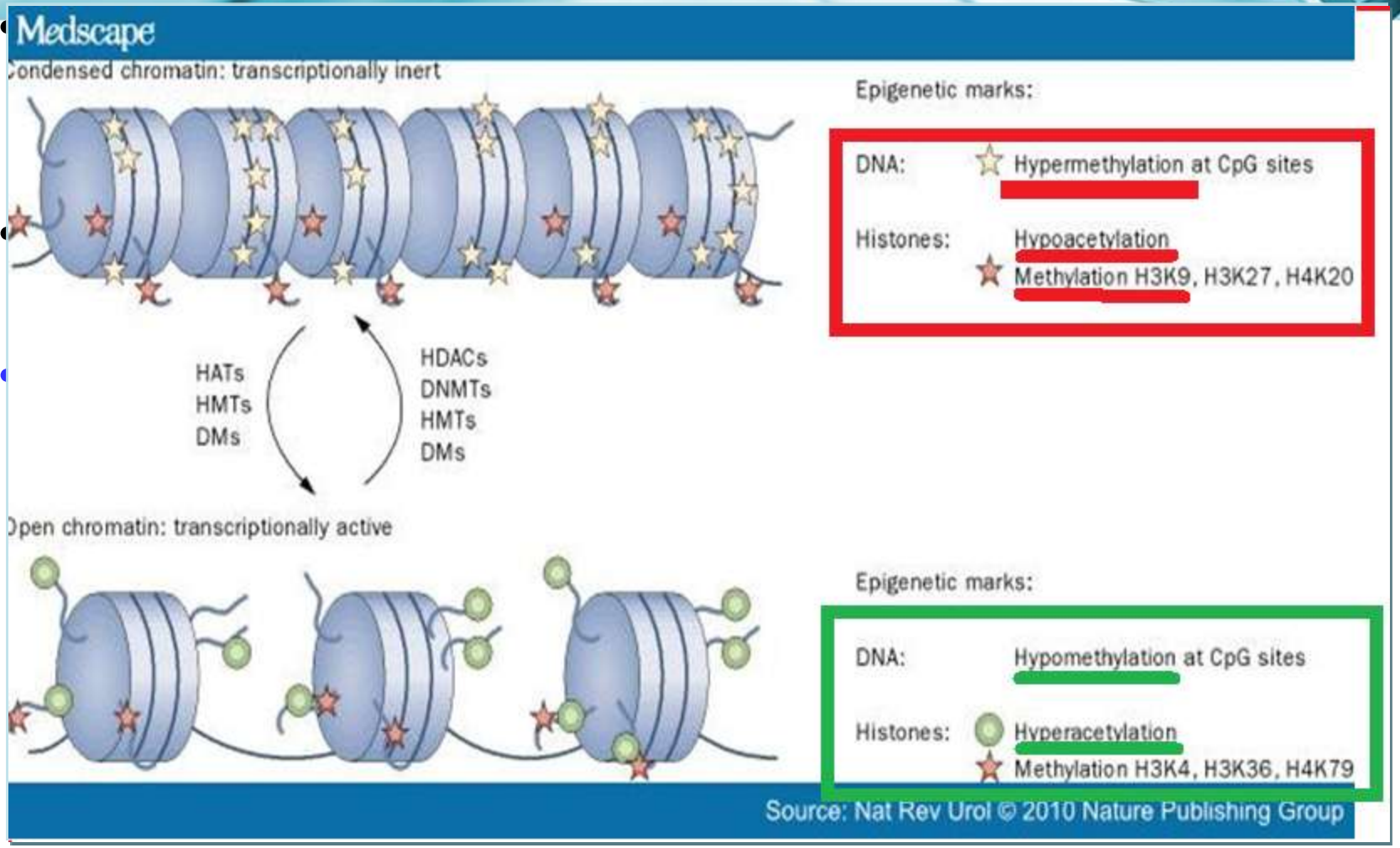


Histone Modifications

- Histone Tails are subject to a variety of covalent modifications by enzymes
  - ***Acetylation***
  - ***Deacetylation***
  - ***Methylation***
  - ***Demethylation***
  - ***Phosphorylation***

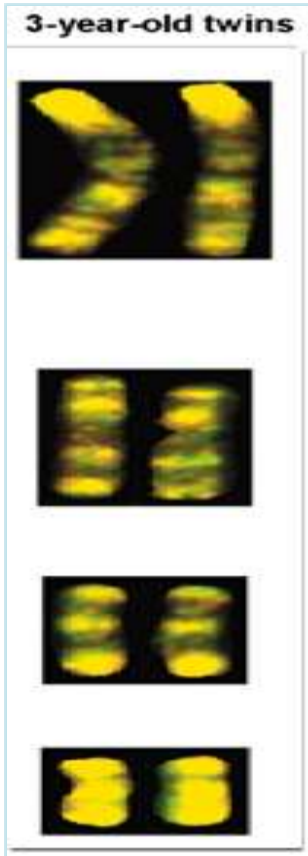


“Histone Code” hypothesis



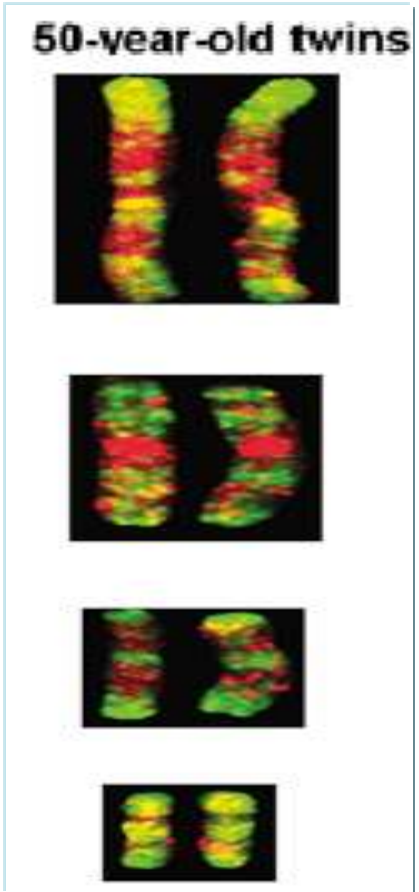
Epigenetic differences arise during the lifetime of monozygotic twins

- Identical twins may have a different metylation pattern
- Epigenetic Factors Can Explain Susceptibility to Illness



very similar distribution of DNA methylation indicated by the presence of the yellow color obtained by equal amounts of the green and red dyes

abundant changes in the pattern of DNA methylation observed by the presence of green and red signals that indicate hypermethylation and hypomethylation events



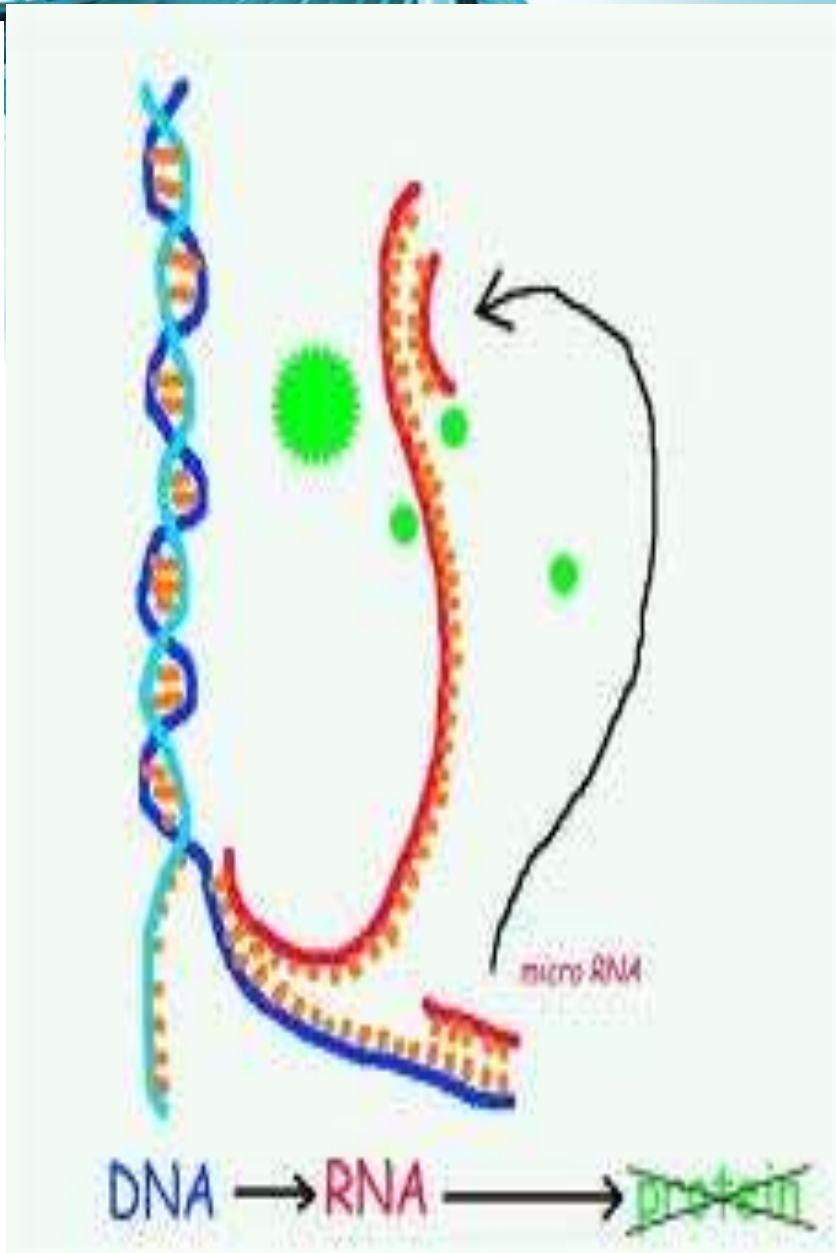
3-Micro RNA(miRNA) or small interfering RNA (siRNA)

- miRNAs are endogenously expressed **18-24 nucleotide** single strand *small non coding RNA molecules* that principally function by binding to the 3' untranslated region of mRNA and repress the expression of their gene products (at transcriptional and post transcriptional levels). ***Lee RC, et al 1993***
- Unlike small interfering RNAs in plants , animal miRNA don't require complete complementarity to bind their targets. Thus one miRNA can regulate *multiple mRNAs* and one mRNA can be repressed by different miRNAS.

He L, Hannon GJ. 2004

- Expression of miRNAs is altered in many pathophysiologic conditions and regulation of miRNAs by drugs is essential for drug activity.

- Several miRNAs circulate in the blood and can be used as biological markers for diagnosis and prognosis of specific cancers, myocardial ischemia, renal and hepatic injury.



Lorenzen JM, Thum T. Clin J Am Soc Nephrol 2012

- **Epigenetic role in medicine & Nephrology**
- **Can epigenetics be manipulated?**

NDT
Nephrology Dialysis Transplantation

Nephrol Dial Transplant (2014) 29: 1821–1827
doi: 10.1093/ndt/gfu026
Advance Access publication 27 March 2014

Full Reviews

Epigenetics: a new way to look at kidney diseases

Pazit Beckerman, Yi-An Ko and Katalin Susztak

Renal Electrolyte and Hypertension Division, Department of Medicine, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA, USA

Nephrol Dial Transplant (2008) 23: 1493–1496
doi: 10.1093/ndt/gfn056
Advanced Access publication 28 February 2008

Editorial Review

Epigenetics—a helpful tool to better understand processes in clinical nephrology?

Peter Stenvinkel[1] and Tomas J. Ekström[2]

[1]Division of Renal Medicine, Department of Clinical Science, Intervention and Technology and [2]Department of Clinical Neuroscience, Karolinska Institutet, Stockholm, Sweden

BMC Nephrol. 2011; 12: 27.
PMCID: PMC3154145
Published online 2011 Jun 16. doi: 10.1186/1471-2369-12-27

Renal kallikrein excretion and epigenetics in human acute kidney injury: Expression, mechanisms and consequences

Sun Woo Kang,[#1] Pei-an Betty Shih,[#2] Roy O Mathew,[#3] Manjula Mahata,[2] Nilima Biswas,[2] Fangwen Rao,[2] Liying Yan,[4] Josee Bouchard,[5] Rakesh Malhotra,[6] Ashita Tolwani,[7] Srikrishna Khandrika,[6] Ravindra L Mehta,[✉6] and Daniel T O'Connor[✉2,6]

Therapeutic use of miRNA antisense may be beneficial, if given early enough, to ameliorate or even stop further injury in AKI and this has been proved in AKI induced by I/P injury in mice.

CKD

Nephrol Dial Transplant (2014) 29: i1–i8
doi: 10.1093/ndt/gft361
Advance Access publication 17 September 2013

ndt
Nephrology Dialysis Transplantation

Full Reviews

Evidence for the involvement of epigenetics in the progression of renal fibrogenesis

Björn Tampe and Michael Zeisberg

Department of Nephrology and Rheumatology, Göttingen University Medical Center, Georg August University, Göttingen, Germany

- HISTONE MODIFICATIONS
- histone acetylation has been best studied in the context of CKD, because histone acetylation can be interfered with by using pharmacological inhibitors of histone deacetylases **(HDACs)** reducing fibrosis. ***Van Beneden K, et al. 2013***

- CPG METHYLATION
- RASAL1 encodes for In experimental kidney injury, acute renal damage and chronic progressive fibrosis are associated with transcriptional repression of Rasal1.
- While spontaneous kidney regeneration after acute injury is associated with normalized Rasal1 expression, Rasal1 expression remains silenced in chronic progressive fibrosis due to Rasal1 hypermethylation.

Bechtel W, et al. 2010

- **DNA HYDROXYMETHYLATION**
- In the context of kidney fibrosis, the role of endogenous DNA demethylation mechanisms in the homeostasis of epigenetic modifications has not been described yet. ***Ficz G, et al. 2011***

- **DNA METHYLTRANSFERASES**
- As hypermethylation can be observed in kidney fibrosis associated with an increase in DNMT1, these mice show ameliorated aberrant promoter methylation.

Bechtel W, et al. 2010

Table 3. Studies of pharmacological modulation of the epigenome in experimental kidney fibrosis

Name	Experimental model	Effect
5-Azacytidine	Nephrotoxic serum nephritis [59]	DNA demethylation
5-Aza-2-deoxycytidine	Uninephrectomy-induced glomerulopathy [88]	*Klotho* demethylation
Dexmedetomidine	Sepsis-induced kidney injury [17]	BMP7 induction
	Radiocontrast-induced nephropathy [89]	Decreased tubular necrosis
	Ischaemia–reperfusion injury [90]	JAK/STAT inhibition
MS-275	Unilateral ureteral obstruction [20]	TGF-beta1/EGFR inhibition
Phenylbutyrate	Streptozotocin-induced diabetic nephropathy [86]	ER stress/p-JNK inhibition
Trichostatin A	Doxorubicine-induced nephropathy [21]	Less macrophage infiltration
	Unilateral ureteral obstruction [16, 22]	Caspase-3 inhibition
	TGF-beta1-induced nephropathy [23]	TGF-beta1/ROS inhibition
	Streptozotocin-induced diabetic nephropathy [23]	TGF-beta1/ROS inhibition
	Nephrotoxic serum nephritis [91]	BMP7 induction
Tubacin	Polycystic kidney disease [24]	EGFR inhibition
Valproic acid	Doxorubicine-induced nephropathy [21]	Less macrophage infiltration
	TGF-beta1-induced nephropathy [23]	TGF-beta1/ROS inhibition
	Streptozotocin-induced diabetic nephropathy [23]	TGF-beta1/ROS inhibition

Table 2. Clinical trials of pharmacological erasure of aberrant methylation listed by the US National Institutes of Health

Name	Target	# Clinical trials	Disease
5-Azacytidine	DNMTs DNA/RNA incorporation	243	Carcinomas Graft-versus-host disease Leukaemias Lymphomas Sarcomas
Decitabine	DNMTs DNA/RNA incorporation	132	Carcinomas Leukaemias Lymphomas Sarcomas
Hydralazine	DNMTs	30	Carcinomas Hypertension Leukaemias

Björn Tampe and Michael Zeisberg, 2013

- **Dialysis patients**
- with renal neoplasm showed evidence of DNA hypermethylation of various genes compared to renal neoplasm cases with normal renal function, it appears that CKD and/or the dialysis procedure per se promotes hypermethylation.
- in the uraemic milieu DNA hypermethylation causes silencing of erythropoietin expression

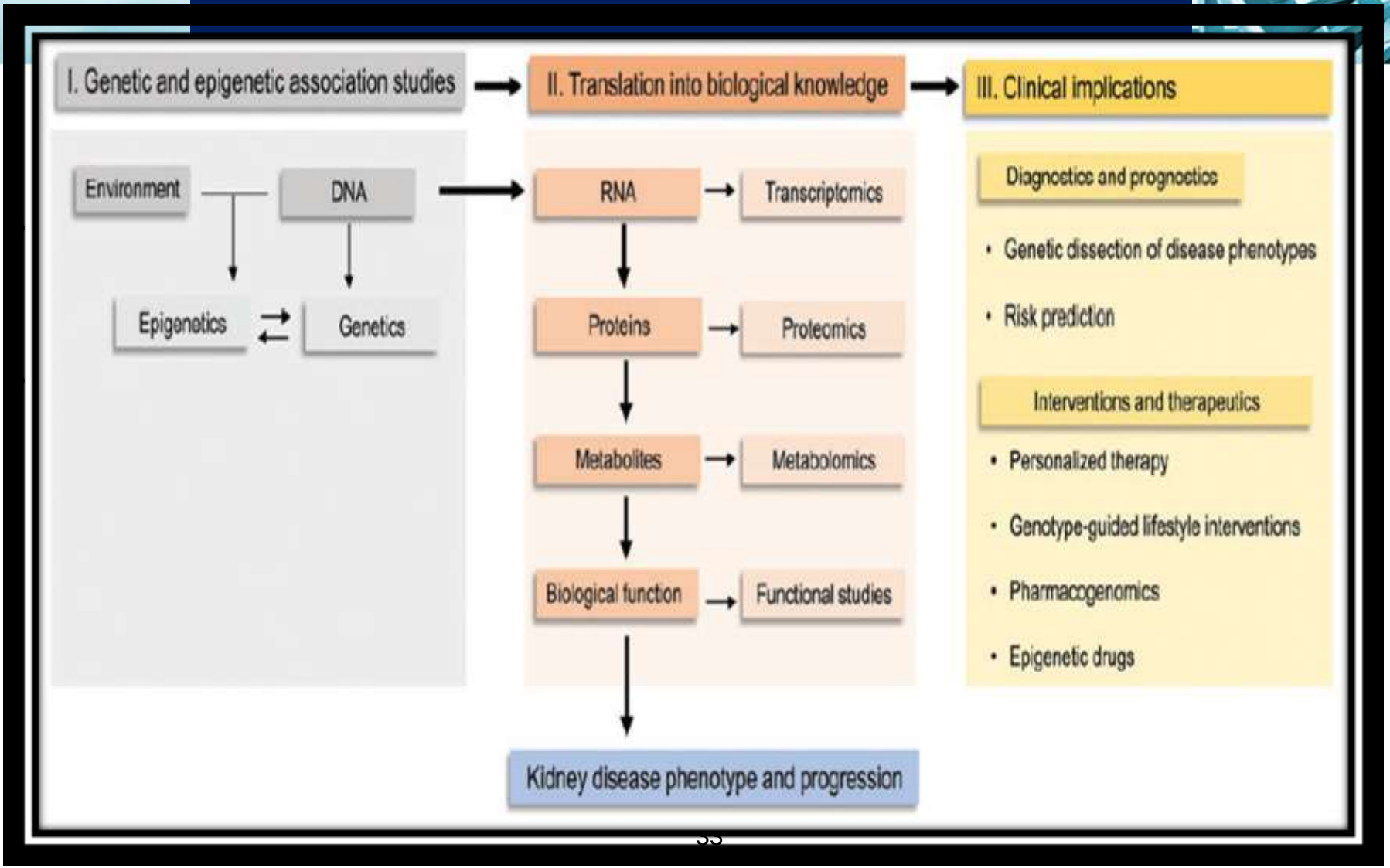
Yin H, Blanchard KL. 2000

Can epigenetics be manipulated?

- In theory, epigenetic drugs could possess higher therapeutic potential and much lower rate of adverse effects in comparison to current treatments, such as DNA methylation inhibitors and histone deacetylase inhibitors, exist at various stages of development.
- Although promising results have been reported, major concerns include ***(i.e. lack of target specificity)*** as well as their transient and long term effects.

Ptak C, Petronis A 2008

Take Home Message




PUGH
SCHOOL EXAMS
FAIL
'It's all your fault, dad – terrible genes'


thank you!